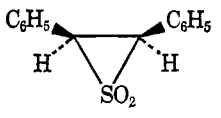
double bond, which is clearly revealed in the ultraviolet spectrum, could not be unequivocally identified in the infrared spectrum presumably because of interfering phenyl absorption.[17] The n.m.r. spectrum of I exhibits only a complex multiplet centered at δ 7.55 due to the aromatic protons.

At its melting point smooth decomposition of I occurs with the formation of diphenylacetylene (72%) and sulfur dioxide. Firm evidence for the presence of



V

the three-membered ring in sulfone I was obtained by reduction of the double bond by means of aluminum amalgam[18] at -45 to $-40°$ in wet ether containing 0.5% ethanol. This gave the corresponding *cis*-episulfone (V),[19] m.p. 85–88° dec., in 8% yield (16% based on recovered I). For purposes of comparison the episulfone V was prepared in 49% yield by a well-known general method[7,20,21] involving treatment of phenyldiazomethane with sulfur dioxide at $-30°$. The stereochemistry of V is assigned on the basis[7] of its decomposition at the melting point to *cis*-stilbene (83%).

Although the question of the possible aromatic stabilization of the vinylene sulfones must await further work now in progress on I as well as on the parent heterocycle and its alkyl and other aryl derivatives, it is striking that the unsaturated sulfone I is remarkably more stable than the saturated analog V. Solutions of V in chloroform at room temperature rapidly developed the odor of sulfur dioxide, and infrared examination showed that in 1–2 days most of the sulfone had undergone conversion to *cis*-stilbene. On the other hand the unsaturated sulfone I appeared to be stable for at least 2 months in chloroform solution at room temperature and in fact could be recrystallized easily from hot benzene.

Acknowledgment. Thanks are due to the donors of the Petroleum Research Fund, administered by the American Chemical Society, and the U. S. Army Research Office, Durham, for partial support of this work.

(15) These tentative assignments are based on the shifts to longer wavelengths (7.91 and 8.62 μ, respectively) which are observed in chloroform solution.[14a,16] The band at 7.82 μ is somewhat outside the range normally quoted[14] for the asymmetric stretching mode, but a less intense band at 7.57 μ, which falls more closely on the Bellamy–Williams internal correlation line,[14b,c] is insensitive to solvent or phase change (carbon tetrachloride, chloroform, Nujol mull, KBr pellet).

(16) S. Ghersetti, *Boll. sci. chim. ind. Bologna*, **21**, 237 (1963).

(17) On the other hand a number of acyclic, phenylated α,β-unsaturated sulfones show a moderately intense band at 6.15 μ due to the conjugated double bond.

(18) F. L. Hahn and E. Thieler, *Ber.*, **57**, 671 (1924).

(19) In contrast to the case of I the infrared spectrum of V shows only two intense bands in the regions normally associated with the SO_2 bands: $\lambda_{max}^{CCl_4}$ 7.43 and 8.62 μ; $\lambda_{max}^{CHCl_3}$ 7.49 and 8.67 μ.

(20) H. Staudinger and F. Pfenninger, *Ber.*, **49**, 1941 (1916).

(21) G. Hesse and S. Majmudar, *ibid.*, **93**, 1129 (1960).

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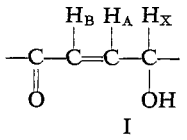
The Isolation and Structural Elucidation of a Novel Steroidal Tumor Inhibitor from *Acnistus arborescens*[1,2]

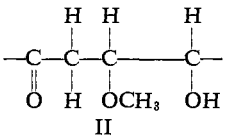
Sir:

The leaves of *Acnistus arborescens* (L.) Schlecht (Solanaceae) and related species have been used for many years to treat cancerous growths.[3] During our search for tumor inhibitors from plant sources, alcoholic extracts of dried *A. arborescens* leaves[4] showed significant inhibitory activity when tested *in vitro* against cells derived from human carcinoma of the nasopharynx (KB) and *in vivo* against sarcoma 180 in mice.[5] We report herein the isolation and structural elucidation of a novel steroidal tumor inhibitor from *A. arborescens*.

Fractionation of the ethanol extract, guided by assay against sarcoma 180, revealed that the active principle was concentrated, successively, in the chloroform layer of a chloroform–water partition, in the aqueous methanol layer of a 10% aqueous methanol–petroleum ether partition, and in the chloroform layer of a 45% aqueous methanol–chloroform partition. Further fractionation involving alumina chromatography yielded compound A,[6] $C_{28}H_{38}O_6$, m.p. 252–253°, $[\alpha]^{28}D$ +125°,[7] which shows λ_{max}^{alc} 214 mμ (ϵ 17,300), λ_{max} 2.81, 2.94, and 5.92 μ (broad), and n.m.r. signals at τ 3.05 (H_A, quartet, J_{AB} = 10 c.p.s., J_{AX} = 6 c.p.s.), 3.82 (H_B, doublet, J_{AB} = 10 c.p.s.), 6.25 (H_X, doublet, J_{AX} = 6 c.p.s.) for the ABX system of I, 5.64 (2 H, $=CCH_2OH$), 6.78 (1 H, epoxy H), 7.95 (3 H, $-C=CCH_3$), 8.58 and 9.28 (6 H, two tertiary CH_3), and 9.02 (3 H, doublet, J = 7 c.p.s., one secondary CH_3). Compound A was



I



II

converted to several crystalline derivatives: the diacetate, $C_{32}H_{42}O_8$, m.p. 201–202°, $[\alpha]^{30}D$ +192°, λ_{max}^{alc} 214 mμ (ϵ 18,000), λ_{max} 5.76 (acetate ester), 5.86 (α,β-unsaturated δ-lactone), and 5.94 μ (α,β-unsaturated ketone); the tetrahydrodesoxy derivative (by hydrogenation with hydrogen and palladium), $C_{28}H_{42}O_5$, m.p 228–229°, λ_{max} 2.79, 5.78 (δ-lactone), and 5.86 μ (ketone); the methanol adduct, $C_{28}H_{42}O_7$, m.p. 242–243°, $[\alpha]^{35}D$ +19°, λ_{max}^{alc} 217 (ϵ 9500), λ_{max} 2.79, 2.94, 5.86 (α,β-unsaturated δ-lactone), and 5.91 μ (ketone), n.m.r. spectrum showing the presence of a methoxyl

(1) Tumor Inhibitors. XIV. Part XIII in the series: S. M. Kupchan, S. J. Barboutis, J. R. Knox, and C. A. Lau Cam, *Science*, in press.

(2) This investigation was supported by grants from the National Cancer Institute (CY-04500) and the American Cancer Society (T-275).

(3) R. de Grosourdy, "El Médico Botànico Criollo," F. Brachet, Paris, 1864; F. Haüssler, *Schweiz. Apoth.-Ztg.*, **52**, 260, 275 (1914). We thank Dr. Jonathan L. Hartwell of the National Cancer Institute for calling these references to our attention.

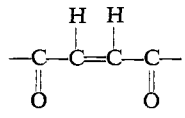
(4) The plant material was collected by J. A. S. R. in Costa Rica in Jan. 1961.

(5) Cytotoxicity and *in vivo* inhibitory activity were assayed, under the auspices of the Cancer Chemotherapy National Service Center, National Cancer Institute, by the procedures described in *Cancer Chemotherapy Rept.*, **25**, 1 (1962).

(6) Compound A showed significant inhibitory activity against sarcoma 180 in mice at 20 mg./kg., and cytotoxicity (ED_{50}) against KB cell culture at 0.15 μg./ml.[5]

(7) All rotations and infrared spectra are in chloroform, unless otherwise noted. N.m.r. spectra were determined on a Varian Associates A-60 spectrometer in deuteriochloroform; chemical shifts are reported in τ values (p.p.m.). Satisfactory analyses have been obtained for products with cited empirical formulas.

group (τ 6.62, 3 H) and absence of vinyl protons, in accord with system II; the dihydrodesoxy derivative of the methanol adduct, m.p. 243–244°, $C_{29}H_{44}O_6$, $[\alpha]^{27}D$ +6°, λ_{max} 2.80, 5.78, and 5.83 μ; the dehydro derivative (by oxidation with manganese dioxide), $C_{28}H_{36}O_6$, m.p. 276–277°, $[\alpha]^{27}D$ +106°, λ_{alc}^{max} 223 mμ (ϵ 14,700),[8] λ_{max} 2.80 and 5.85–5.91 μ, n.m.r. spectrum showing the absence of ABX system of 1 and a new sharp signal at τ 3.20 (2 H), in accord with system III;



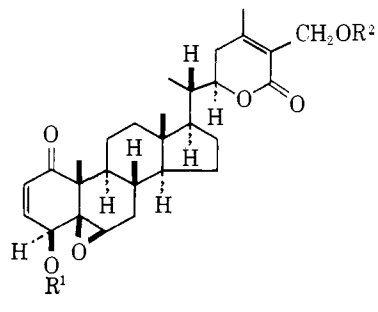
III

the *p*-bromobenzoate, $C_{35}H_{41}BrO_7$, m.p. 196–197°, $[\alpha]^{28}D$ +63°; λ_{max}^{Nujol} 2.85, 5.85, 5.92, and 13.20 μ; and the *p*-bromobenzoate monoacetate, $C_{37}H_{43}BrO_8$, m.p. 179–180°, $[\alpha]^{28}D$ +101°, λ_{max}^{Nujol} 5.73, 5.84, 5.92, and 13.15 μ.

The *p*-bromobenzoate monoacetate crystallized from ethyl acetate in the orthorhombic system, space group $P2_12_12_1$, with cell dimensions a = 14.74, b = 40.35, c = 12.95 Å. We found the crystal density to be 1.276 g. cm.$^{-3}$, indicating that if the molecular formula of the *p*-bromobenzoate monoacetate is $C_{37}H_{43}BrO_8$ then the asymmetric crystal unit consists of two molecules of $C_{37}H_{43}BrO_8$ and one molecule of ethyl acetate (D_{calcd} = 1.276 g. cm.$^{-3}$); the crystallographic problem therefore involved the location of 96 independent carbon and oxygen atoms and 2 bromine atoms.

The initial positions of the two independent bromine atoms were derived from the three-dimensional Patterson synthesis and the carbon and oxygen atoms were then located by evaluating three-dimensional electron-density distributions with Fourier coefficients weighted according to the method proposed by Sim.[9] Three Fourier syntheses were used in the elucidation of the structure, and the approximate atomic coordinates are now being refined by the least-squares method. The present value of R for 3427 independent X-ray reflections is 18%. A most important verification of the structure lies in the fact that we find two crystallographically distinct but chemically identical molecules.

Our results establish that the *p*-bromobenzoate monoacetate has the structure and stereochemistry IV, R^1 = Ac, R^2 = *p*-BrC_6H_4CO, from which it follows that compound A is represented by IV, R^1 = R^2 = H.



IV

(8) *Cf.*, *e.g.*, the homocisoid enedione described by D. H. R. Barton, *et al.*, *J. Chem. Soc.*, 2988 (1951); 2210 (1952).

(9) G. A. Sim, *Acta Cryst.*, **12**, 813 (1959); **13**, 511 (1960); "Computing Methods and the Phase Problem in X-ray Crystal Analysis," R. Pepinsky, J. M. Robertson, and J. C. Speakman, Ed., Pergamon Press, Oxford, 1961, p. 227.

While this work was in progress, Lavie, *et al.*, reported the isolation[10] and partial elucidation of the structure of withaferin A[11] from *Withania somnifera* Dun. The similarity in physical properties of compound A to those reported for withaferin A led to direct comparison of the respective materials and to demonstration of their identity (by mixture melting point, mixed t.l.p.c., and infrared studies).[12,13] Consequently, we adopt the name withaferin A for IV, R^1 = R^2 = H.

(10) A. Yarden and D. Lavie, *J. Chem. Soc.*, 2925 (1962).

(11) D. Lavie, E. Glotter, and Y. Shvo, *Israel J. Chem.*, **2**, 247 (1964); *J. Org. Chem.*, **30**, 1774 (1965).

(12) We thank Professor David Lavie cordially for comparison samples of withaferin A and of withaferin.[10] Withaferin was found to be identical with the methanol adduct of withaferin A (see above).

(13) NOTE ADDED IN PROOF. Professor Lavie recently has informed us that his structural studies have led independently to proposal of the same structure for withaferin A: D. Lavie, E. Glotter, and Y. Shro, *J. Chem. Soc.*, in press.

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Free Radical Additions of Carbon Tetrahalides to *cis*-Cyclooctene[1,2]

Sir:

The appearance of brief reports of evidence for a homolytic transannular reaction competing with normal 1,2 addition of bromotrichloromethane to methylenecycloalkanes[3] prompts us to make a preliminary communication about our study of additions of carbon tetrahalides to cyclooctene, a study which firmly establishes the occurrence of transannular hydrogen atom transfer during addition to an eight-membered ring and substantially delineates the energy requirements for the process.

Both photo- and thermally initiated[4] additions of carbon tetrachloride to *cis*-cyclooctene[5] give mainly isomeric $C_9H_{14}Cl_4$ products.[6] In contrast to a patent claim that the thermal addition gives a 1,2-addition product,[7] we found that at least 97% of the $C_9H_{14}Cl_4$

(1) Acknowledgment is made to the donors of the Petroleum Research Fund, administered by the American Chemical Society, for partial support of this research (Grant 1817-A4).

(2) Presented at the 150th National Meeting of the American Chemical Society, Atlantic City, N. J., Sept. 13, 1965; Abstracts, p. 6S.

(3) (a) For addition to methylenecyclodecane, see M. Fisch and G. Ourisson, *Chem. Commun.* (London), 407 (1965). (b) For addition to the more rigid olefin, longifolene, see G. Ourisson, *Proc. Chem. Soc.* (London), 281 (1964).

(4) Photoinitiated additions were carried out in quartz flasks suspended in a Rayonet photochemical reactor and irradiated at 44° for 40 hr. with 2537 Å. light. Thermally initiated additions were accomplished without added initiator in a Parr medium pressure apparatus heated at 155° for 4.5 hr. Both reactions utilized about 4 moles of carbon tetrachloride per mole of cyclooctene.

(5) *cis*-Cyclooctene was generously given to us by Columbian Carbon Co., Lake Charles, La.

(6) Satisfactory analyses were obtained for all new compounds mentioned in this communication.

(7) F. Reicheneder and H. Suter, German Patent 1,036,847 (August 21, 1958); *Chem. Abstr.*, **54**, 22416c (1960).